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BACTERIA IN CHEESE.*

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Within the last few years the literature on our subject has accumulated at a rapid rate so that a complete review of the work done and results attained would be out of place here; but a brief summary of the historical aspect together with a statement of the present status of the problem is of interest to most readers.

Up to the time of Pasteur's memorable studies in fermentation, the latter process was universally considered as a purely chemical phenomenon having no relation to organized life. His convincing results opened the way to many new fields of investigation, among which was the curing of cheese. The latter being considered a species of fermentation was naturally explained as due to organisms. Besides this Pasteur had proved that the lactic fermentation or souring of milk was due to bacteria. What could be more plausible than that the changes occurring in ripening cheese should be due to similar causes?

Cohn,[1] who fell under the influence of Pasteur's work, reopened the cheese problem at the point where the chemists had left it, by demonstrating bacteria in the rennet used in cheese-making, and concluded on this meager basis that the process was due to bacteria. But the first real work in our field, which has now become classical, was done by Duclaux, an assistant of Pasteur's and who was, beyond doubt profoundly

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[1] Cohn, Ferdinand. Beitrage zur Biologie der Pflanzen, Bd. I, 3 Heft. p. 191.

influenced by Pasteur's great discoveries in the field of fermentation. Duclaux[1] in 1878, lent support to Cohn's conclusion in that he found that cheese made from sterilized milk failed to ripen. This certainly appeared as conclusive proof that the phenomenon in question was due to germs, although it is now certain that his results were due to quite different causes.

Soon other experimenters followed in this newly opened field, whose results quite uniformly lent support to the fermentation theory.

Swiss or Emmenthaler cheese was examined microscopically by Benecke[2] and was found to contain a certain bacillus especially abundant during the ripening stage, which he considered as the particular germ that ripened the cheese. About the same time methods for isolating germs and growing them in pure cultures were being discovered, and Duclaux,[3] making use of these, succeeded in isolating ten species of bacteria from cheese, seven of which he found to be aerobes, and three anaerobes. He designated these as Tyrothrix forms, because of their occurrence in chains or threads. All these germs were peptonizers, in that they liquefied gelatin and digested the casein of milk. A peptonizing ferment was also isolated from the products of their growth, which changed the casein of cheese into soluble peptones. Thus his theory appeared established on a secure foundation.

However the above work antedates the discovery of solid media such as gelatin and agar, and a doubt may be expressed as to the validity of this work. From our present knowledge of cheese bacteria, it appears quite remarkable that all these separate forms should have such similar characteristics, and that none of the non-peptonizers were discovered. True, his meth-

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[1] Duclaux, Fabrication, maturation et maladies du fromage de Cantal. Annales agromomiques 1878.

[2] Benecke and Schulze, Untersuchungen über den Emmenthaler Kase und über einige andere schweizerische Kasesorten. Landw. Jahrbucher XVI, pp. 317-400.

[3] Duclaux, Le Lait, etudes chemiques et microbiologiques. Paris 1887. (Deuxieme tiragu augmenta 1894.)

ods were not adapted to the isolation of the latter class but we now know that peptonizers are not abundant nor are the species numerous in most kinds of cheese. Hence it is quite possible that several, if not most of Duclaux' species, were duplicates of each other.

For nearly a score of years little progress was made. A more accurate knowledge of the chemical products of the curing process was obtained; also a better knowledge of the bacterial flora. From the latter point of view, Adametz'[1] work deserves especial mention, for he isolated nineteen species of bacteria from Emmenthaler cheese—six micrococci, five sarcina and eight bacilli. These different species were described, and their physiological activity noted. He found too, that soft cheese contained a greater numerical percentage of germs than hard varieties; that the number increased as the cheese ripened, reaching as high as 850,000 per gram of cheese. Where disinfectants were added to the cheese the germ content decreased and ripening proceeded more slowly or ceased entirely. This latter observation the writer has not been able to corroborate and remains quite inexplicable in the light of our present knowledge.

Of yet greater importance was the able work of Von Freudenreich,[2] who also investigated Emmenthaler. He isolated a number of species, and made quantitative determinations of the germ content. Earlier investigators had found numerous peptonizing bacteria, with relatively few non-preptonizing forms. Von Freudenreich found on the contrary, that the former class was relatively small in numbers and belonged to the classes of Hay and Potato bacilli; the latter class predominated and frequently constituted the only bacteria present. These he found to belong to the class of lactic-acid-producing germs. Thus it appeared that if cheese was ripened by bacteria, the former theory was no longer tenable; that casein digesters took

[1] Adamatz Bakteriologisch Untersuchung über den Reifungs prozess der Kase. Laudw. Sahrbucher XVII, 1897, pp. 227–269.

[2] Von Freudenreich, Weitere bakteriologische Untersuchungen über den Reifungs prozess des Emmenthaler Kases. Land. Jahrbucher des Schweiz. VIII, 1894, pp. 207–239.

no part in the curing process, but that this function must be ascribed to the non-peptonizing lactic-acid—producing germs.

To this, however, he was loathe to subscribe, for how could the acid-producing germs which did not affect the casein in milk cultures, except to precipitate it, produce such profound changes in the cheese? New media and methods were tried to develop bacteria which had perhaps been overlooked or lost entirely. In these attempts one or two anaerobes were found, but no important phenomena could be connected with them.[1]

Besides the above a number of other investigators had contributed data to our problem, but none which advanced materially toward an explanation of the ripening process. Pammel[2] had discovered an aromatic bacillus in cheese, while Russell[3] had isolated gas germs which caused "huffing" or swelling of cheese. Lloyd[4] of England claimed to find B. acidi lactici in English cheddar.

This was essentially the situation when the writer, then a student under Dr. H. L. Russell of the Wisconsin University and Experiment Station, first took up the problem. Some of the observations had been made by means of a microscopical examination of cheese; others had been carried out before the time of solid media; the most recent labors had been accomplished without uniform or established methods and while tending to overthrow proposed theories afforded no solution to the question.[5]

Our first attempt was to find a suitable method for isolating the bacteria and at the same time to obtain reliable quanti-

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[1] Von Freudenrich, Forschung auf den Gebiete des Kasereifungsprozesses. Cent. fur Bakt. II Abt. I 1895, No. 17, 19.

[2] Pammel, An Aromatic Bacillus of Cheese. Bul. No. 21, Iowa Exp. Station. Ripening of Cheese. Ibid.

[3] Russell, Gas-producing bacteria and the relation of the same to cheese. 12th An. Reports Wis. Exp. Station 1896, p. 139.

[4] Lloyd, Observations on Cheddar Cheese-making. Bath and W. Eng. Soc. Reports for 1891, 2 and 3.

[5] Russell and Weinzirl, Cent. f. Bakt. II Abt. Bd. 3, pp. 456-467, 1897.

tative data. This was no easy task, for the cheese had to be brought into solution or finely divided so as to thoroughly separate the germs. The latter course was the only one which seemed feasible. The cheese could not be satisfactorily triturated in water, so sterilized sand and finally sterilized sugar was tried. Although not entirely satisfactory, the sugar readily dissolved in the water in which the dilutions were made. A gram of cheese was taken as the unit of weight, and all vessels and instruments used were thoroughly sterilized. The glassware and media were sterilized by heat; the cheese-trier, mortar, etc., by flaming; and the sugar by the addition of ether for one or more days. Heat was not used for the latter purpose for it tended to caramelize the sugar or even melt it when the temperature rose above 140° C. The ether gave very satisfactory results and could be easily applied.

Some time was spent in preliminary experiments and testing of methods after which a cheddar cheese (Flat) was made in the University Dairy School and analyzed bacteriologically at suitable intervals. For this purpose ordinary neutral peptone gelatin was used, although other modifications of gelatin and agar were employed as checks upon the work. No material advantages were gained by the latter, however.

Petri dishes were used in these analyses and three plates, using appropriate dilutions as suggested by experience and the age and condition of the cheese, were regularly made. These were allowed to develop from five to ten days and the colonies counted and pure cultures made. New cheese were made and similarly treated. A great amount of time was absorbed in these laborious countings, computations, and making of cultures, the results of which, for the sake of simplicity are tabulated. See page 154.

It will be seen from an inspection of the table, that the germ content of our cheese was very much higher than any figures reported by previous investigators, reaching at times scores of millions. Whether these results are due to improved methods of analysis or to differences in the kind of cheese, we

Bacteria in cheese. — Table of num

TABLE I.

No. of Bacteria per gram in Cheddar Cheese at different stages of the Ripening Process.

	Age of cheese	Total no. bacteria pr. gr.	Lactic acid bacteria pr. gr.......	Gas producing bacteria pr. gr.....	Casein digesting bacteria pr. gr....	Inert bacteria pr. gr.....	Percentage of lactic acid bacteria in sample....
Cheese I	10 c c milk	53,180,000	40,068,000	6,070.000	6,330 000	100,000	76.5
made 18, VI.	5th day	Analytical	results lost	owing to	liquefac-	tion of	gelatin
1895. Rip-	13th day	68,015,000	67,470,000	400,000	145,000	------	99.2
ened at 18° C.	24th day	69,485,000	69,270,000	210,000	5,000	------	99.3
	36th day	16,996,000	16,996,000	34,000	2,000	------	99.7
	52d day	11,500,000	11,470,000	25,000	----------	------	99.7
	74th day	6,700,000	6,682,000	9,000	----------	------	99.8
	94th day	4,183,000	4,158,000	25,000	----------	------	99.4
	120th day	2,352,000	2,352,000	---------	----------	------	100.
	155th day	207,000	207,000	---------	----------	------	100.
	183d day	380,000	380,000	---------	----------	------	100.
	197th day	377,000	377,000	---------	----------	------	100.
	237th day	86 000	86,000	---------	----------	------	100
Cheese II	10 c c milk	95,000,000	82,600,000	6,030,000	6,180,000	190,000	84.8
made 10. VII	1st day	7,644,000	6,103,000	959,000	581,000	------	79.
1895 ripened 20° C.	5th day	95,640,000	94,300,000	1,243,000	----------	------	98.6
Cheese III	10 c c milk	262.000,000	52,400,000	78,600,000	131,000,000	------	20.
made 24, IX	4th day	115,400,000	115.130 000	271,000	----------	------	99.8
1895, ripened	10th day	64,350,000	64,286,000	64,000	----------	------	99.9
at 20°—24°C.	18th day	43,264,000	43,259,000	5,000	----------	------	99.9
	30th day	36,887,000	36,882,000	5,000	----------	------	99.9
	53d day	5,304,000	5,299,000	5,000	----------	------	99.9
	86th day	15,223,000	15,210,000	---------	----------	------	99.9
	108th day	7,084,000	7,080,000	---------	----------	------	------
Cheese IV	4th day	110.146.000	110,136.000	10,000	----------	------	99.9
Pasteurized	10th day	53,976,000	53.976,000	trace	----------	------	100.
milk with lac-	18th day	38,842,000	38,842,000	---------	----------	------	100.
tic acid made	30th day	23,400,000	23,400,000	---------	----------	------	100.
24, IX, 1895	53d day	1,075,000	1,072,000	3,000	----------	------	99.7
ripened at 20°	86th day	21,000	21,000	---------	----------	------	100.
—24°c.	108th day	5,000	5,000	---------	----------	------	100.
Cheese V.	10 c c milk	665,220,000	664 600.000	---------	----------	620,000	99.9
Normal	Curd	158,320,000	156.000.000	---------	2,320,000	------	99.8
milk and sour	2d day	82,414,000	82,134,000	---------	280,000	------	99.6
milk starter	4th "	95,200,000	94,770,000	---------	430,000	------	99.7
made 14 XII	7th "	102,750.000	102.750.000	---------	trace	------	100.
1895, ripened	10th "	84,617,000	84,617,000	---------	trace	------	100.
20°—23° C.	13th "	24,024,000	24,024,000	---------	----------	------	100.
Cheese VI	10 c c milk	2 7,800.000	211,680.000	1,180,000	14,800,000	200,000	92.8
Normal	Curd	26,532,000	22,560,000	180,000	3,792,000	------	85.
milk made 26	1st day	21,060,000	20,176.000	52,000	832,000	------	95.9
II, 1896 ri-	5th day	43,716,000	39,680,000	36,000	4,000,000	------	90.8
pened at 18°	7th day	46,440,000	45,760,000	80,000	600,000	------	98.5
C.	10th day	98,080,000	97,280,000	160,000	640,000	------	99.1
	13th day	97,240,000	96,800,000	176,000	264,000	------	99.5
	15th day	76,400,000	76.000,000	200,000	200,000	------	93.4
	19th day	48,139,000	48,000,000	77,000	62,000	------	99.6
	22d day	16,000,000	15,633,000	349,000	----------	18,000	97.7
	36th day	11,171,000	11,000,000	158,000	13,000	------	98.3

are unable to say. It is to be noted, however, that the Swiss Emmenthaler and the American Cheddar both belong to the same general class of firm cheese. That the numbers should be high in our analysis of milk and cheese in its early ripening stages is not so surprising, when we consider that all the mechanical steps in the manufacture of cheese are such as to offer the most favorable conditions for development of bacteria. The milk is ripened at nearly blood heat and the curd is also developed at a comparatively high temperature.

The most striking fact of our analyses is the overwhelming preponderance of the class of germs we have designated as the lactic acid bacteria, while the casein digesters are relatively few in numbers, and soon disappear from the cheese entirely. This practically confirms the conditions found by Von Freudenreich in Swiss cheese. The preponderance of acid-producing germs is readily explained, in that the art of cheese-making consists largely in producing those conditions which favor the development of these very germs to the exclusion of others. It is the cheese-maker's constant care to produce sufficient acid to make the curd "string on the hot iron," and it is these germs which change the sugar of the milk into the highly desirable lactic acid. Naturally, the question will be asked, what is the use of this acid?

At first thought, it would seem that the curing of the cheese is causally related to this acid or to the acid germs. Von Freudenreich,[1] in a recent article so concludes. By adding chalk to sterilized milk, which was inoculated with a pure culture of a lactic germ, the acidity of the milk was neutralized and the germ could endure for a much longer time than in cultures without the addition of this material. It was found that under these conditions, which he assumes to be parallel with those found in cheese, a certain amount of breaking down of the casein took place, but less than occurs in the cheese normally. This evidence, though strong, is not conclusive, for

[1] Cent. f. Bakt. 2nd. Abt. Bd. III, p. 234.

why should there be this difference between his culture and normal cheese? And when we seek an explanation of the curing process, the case is not at all clear. The ripening consisted in the conversion of the insoluble casein into soluble peptones or allied products. That the acid is not produced by the activity of the germ upon the casein, can scarcely be questioned. Its only source is in the milk sugar. Is the acid produced, then, to react upon the casein and break it down? No such action is known to chemists, but what are the facts? To test this question a special experiment was devised. Lactic acid was added to curd made from pasteurized milk, and the same was made into cheese. No evidence could be obtained that the acid produced the change in question. Can there be a difference in action between commercial lactic acid and that produced in the cheese, especially in the nascent state?

The problem was then attacked in another way. If the acid production in the cheese could be inhibited, would the process of curing also be inhibited? To test this ether, chloroform, etc., were added to newly made cheese and the same were kept in an atmosphere saturated with these anæsthetics. These cheese cured, apparently, quite normally. Bacteriological analyses revealed bacteria, but in greatly reduced numbers—too small to account for the curing. That bacteria were present is readily explained, for they were not killed by the anæsthetics but their vital activities were inhibited. But the breaking down of the casein and curing of the cheese still remains to be explained. If the bacteria are inhibited in their vital activities, then no acid can have been produced to affect the casein. But the casein underwent its characteristic change. Plainly there is an unknown factor which is the cause of this phenomenon. That factor is not related to bacterial life.

It was at this stage of the problem that Drs. Babcock and Russel[1] sought to apply chemical tests to determine the unknown factor. It is here that their brilliant work in isolat-

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[1] Babcock and Russell, Unorganized ferments of milk: a new factor in the ripening of cheese. 14th Rept. Wis. Agrl. Stat. 1897, p. 161.

ing an unknown ferment from milk, was accomplished. They demonstrated conclusively that this unorganized ferment is not produced by bacterial activity, but is inherent in the milk itself. This ferment was found to have the power of changing casein into soluble peptones under asceptic conditions. It was further found that heat destroyed this ferment and this explained the fact why Duclaux was unable to ripen cheese made from sterilized milk, which experiment we had repeated and found to be correct. From their experiments they incline to the belief "that the ripening of hard cheese, instead of being due solely to bacteria, is caused by the joint action of both organized (bacteria) and unorganized ferments (enzymes). The breaking down of the casein is undoubtedly due, in larger part, to the action of enzymes." Thus, after half a century the pendulum has swung back to the side of the chemists. However, the new theory agrees with that of Ducleaux, in so far that both are based upon the action of enzymes; the differe nce being as to their origin. The former considered them bacterial, the latter finds them to be inherent in the milk.

However, to explain the cause of the curing phenomenon, is not the only nor even the essential part of this paper. There still remains to be explained the fact of the enormous numbers of these lactic germs and the relatively few germs of the other types. That the inert forms should be few or absent is what might have been expected. Undoubtedly they gained entrance from the air; but falling in a medium not especially adapted to them, they were crowded out or, so to speak, smothered, by the growth of the acid forms. The absence of the casein digesters is readily explained by the fact that they are inhibited by the activity of the lactic acid organisms. If the latter class is destroyed as in pasteurization, the digesters, having a free field, develop in abundance.

As to the gas germs, it is a fact that these are, as a rule, also moderate producers of acid. That they are correspondingly more numerous and stand next in number to the purely acid forms, is readily understood. But these same gas germs are the bane of the cheese-maker, and how he is able to keep

them down in his product is still a question. That he frequently fails to prevent their development, much of our product bears a sad testimony. That the gas produciug germs are largely associated with filth and general uncleanliness in the dairy, is quite well established. This enables us to apply a ready and all desirable remedy, namely, to remove all traces of filth in all stages of the work. "The Wisconsin curd test"[1] gives still other means of combatting this evil, for by it the sources of the infection can be readily traced.

Apparently the casein digesters may be at times quite harmless, but many of them induce fermentations resulting in bitter by-products which are very undesirable and undoubtedly prevent the product from reaching the highest grade, if indeed, it is not ruined by them. A more detailed study of the various germs of this class is necessary before we can ascribe to them their due importance in the manufacture of cheese.

However, we have still to explain the presence of the acid germs in such enormous numbers. It is quite certain that they are not concerned, at least to any great extent, in the changing of the indigestible casein into the digestible products. Their function as acid producers is no doubt important in communicating to the ripened product that peculiar flavor, so indescribable but so highly desirable, and which lends to cheddar cheese its own peculiar qualities. If this is true, these germs should be found in all cheddar cheese, no matter in what section of the country it is made. So far our investigations were largely limited to cheese made at the Wisconsin Dairy School. We now directed our study to the state at large and finally to other states, to determine the presence of these germs and at the same time to make as detailed a study of the cheese flora as possible. The results of these investigations so far as they related to cheddar cheese are tabulated below:

[1] Babcock, Russell and Decker,— Factory tests for milk. Bul. 67, Wis. Agrl. Exp. Station.

Bacteria in cheese - Table of numbers

TABLE II.

Showing the relative percentages of the different classes of germs in cheese from different states.

No. of cheese	Where made	No. of species	Lactic acid Bacteria %	Gas producing Bacteria %	Casein digesting Bacteria %	Neutral Bacteria %	Remarks.
1	Patch Grove, Wis.	4	Predominated	Present	Present	?	Plates partly liquified. Computations not made Age of cheese 6 days
2	" " "	2	58	42	------	------	Age of cheese 22 days
3	" " "	3	80	3	17	------	Age 18 days
4	" " "	2	75	25	------	------	Made from gassy milk. Age 32 days
5	?	3	50	48	------	2	Cheese "off flavor"
6	Avoca, Wis.	3	37	54	9	------	"Off flavor"
7	Hortonville, Wis.	3	18	82	trace	------	3 mos. in cold storage. Fine cheese
8	Plymouth, Wis.	3	53	47	"	------	Gassy cheese
9	Manitowac, Wis.	4	present	pr's'nt	trace	pr's'nt	Computations not made
10	Plymouth. Wis.	4	27	70	trace	3	Gassy and "off flavor"
11	Plain, Wis.	4	66	33	trace	1	"Off flavor"
12	Trim Belle, Wis.	4	98	------ 1	trace	1	Age 51 days. Fine cheese
13	Cayenovia, Wis.	3	83	13	4	------	Pinholey and bad odor
14	" "	3	95	4	1	------	
15	Wisconsin.	1	100	------	------	------	Age 15mos. 4,030 germs per gram. Lowest no. found
16	Binghamton, Wis.	3	not numerous	many	trace	------	Computation not possible
17	Verfkind, Wis.	3	present	pr's'nt	pr's'nt	------	Plates partly melted. No count
18	Boving, Wis.	1	"				Plates melted. Yeast also present
19	Wisconsin.	4	90	10	trace	------	Age 8 days
20	Hortonville, Wis.	4	57	38	1	4	
21	Wisconsin.	3	23	75	2	------	Age 12 days. Quality not noted
22	" "	2	48	51	------	------	Age 2 wks. Yeasts=1% Quality not noted
23	Tonet, Wis.	3	82	17	------	1	Very fine cheese
24	Peebles, Wis.	3	72	27	1	------	Slightly "off flavor"
25	Ahnapee, Wis.	3	57	42	1	------	Age 22 days
26	Rio Creek, Wis.	3	70	30	------	------	Age 22 days
27	Illinois.	3	82	18	------	------	
28	Ontario, Can.	2	29	17	------	------	Yeast=54%. Yeast assimilated properties of acid germs
29	Minnesota.	5	85.7	10.5	3.5	0.3	
30	New York State.	3	99	0.5	0.5	------	
31	Michigan.	2	39	61	------	------	Poor flavor
32	" "	4	40	59	1	------	Fine cheese
33	Dakota.	4	63	22	5	11	Fine cheese. 2 mos. old
34	Pennsylvania.	3	95	5	trace	------	
35	Meadville, Pa.	3	98	1	------	1	
36	Colorado.	3	100	trace	------	trace	

Before proceeding to a consideration of our data, it should be stated, that the samples were obtained by the use of a cheese borer or trier, the core of cheese placed in a sterile glass test-tube, and this was sent to the Laboratory in a wooden case, usually by mail. Through the kindness of Mr. J. W. Deeker of the Wisconsin Dairy School, most of the samples were so obtained, together with many important data. In all cases, one or more days intervened between the time of sampling and that of analyzing the sample. Although the exterior of the plug was carefully removed by means of a sterile knife, and only the center used, yet it is possible that the normal distribution of the cheese bacteria may have been altered in some degree by the removal of the sample from its natural conditions and the consequent drying. Samples 33-36, however, were obtained directly from fair sized portions of the cheese. It would perhaps have been fortunate, if more of the samples had been so taken, so as to admit of a comparison.

Usually three peptone gelatin Petri plates were made, using three dilutions of the cheese. The first contained, approximately, ½ gr. of cheese added to the tubes of gelatin and finely triturated by means of a sterile glass rod; the second contained from 3 to 5 drops taken from the first; and the third 3 to 5 loopfuls taken from the second. In this way a wide difference in seeding was obtained, and one of the three plates usually presented favorable conditions for counting regardless of the age of the cheese. In old cheese, colonies were usually limited to plate No. 1. New cheese gave the most favorable results on plate No. 3, etc. A special advantage in this method lay in the fact that moderately scarce germs, which would have been missed in Nos. 2 and 3 were brought out in No. 1. This was particularly the case with the liquefiers or digesters, of which there was usually a trace on plate No. 1, but none on the other two plates.

Table II comprises 36 analyses of cheese, 26 of which were Wisconsin product and 10 from other states—a total of nine states being represented, which included a wide range of territory, extending from New York and Pennsylvania on the east to Colorado on the west and Canada on the north. Within this

area is included the cheese belt of our country, and although the analyses are not as numerous as could be desired, they are sufficiently so, to show at least, the main characteristics of the cheese flora. In this work 13 species of bacteria were isolated and grown in pure cultures. These do not include single colonies found on the plates, which we are inclined to believe should not be considered as forming a part of the true cheese flora, but rather as accidental, possibly gaining admmission in the manipulation of the analysis. At any rate no special importance could be attached to such rare colonies, granting that the germs may have existed in the cheese. It is conceivable that all the bacteria of the air and even of water, might be found in cheese, but we do not consider these an integral part of the cheese flora proper.

Of these species, of which the limits of this paper will not permit a description, the lactic acid producing germs are found to be present in all the analyses, and furthermore are the most numerous class in three-fouths of the cheese. The germs producing gas were present in all samples with one exception, (No. 15) and were most numerous in approximately one-fourth of the cheese. In three of these cases (Nos. 6, 10 and 31) the preponderance of gas germs was associated with "off flavors" in the cheese; in two cases, (Nos. 21 and 22) no note had been taken, while in two, (Nos. 7 and 32) the cheese were apparently fine, one of which (No. 7) had been kept for a number of months in cold storage which may have exerted a repressing influence on the multiplication of the lactic organisms.

The bacteria affecting the casein of milk were entirely absent in one-third of the samples, while the germs of a neutral action were present in one-third of the cases. These facts are quite in harmony with our former observations, and while not as striking in many respects, yet remembering the wide area of our field and the varied character and quality of our product, the figures are, on a whole, as uniform as could have been expected. The presence in relatively small numbers or entire absence of the digesting and neutral classes, confirms our conclusion that these play no important function in the cheese and

are present in it only because they were present in the milk, perhaps by accident or carelessness in handling the product, but never really flourish in the cheese medium.

The well-nigh universal distribution of the gas germs is striking and important. That these bacteria are a serious detriment, the cheese-maker and numerous experiments bear convincing testimony. Although the above data were not obtained with any special reference to this subject, yet in most cases we find bad flavors associated with a high content of gas organisms. Their universal presence is explained, perhaps, in that most of these species are also acid producers, hence they find a favorable medium in the cheese. From an economical aspect, to overcome these germs is a very important problem on which considerable labor has been spent. The use of pure cultures of lactic organisms, in one form or another, has been recommended and if practiced in accordance with bacteriological methods, would undoubtedly be of considerable help; but preventative measures should also be adopted. Pure cultures cannot overcome the effects of slovenliness in any of the departments of the dairy. Fortunately, the Wisconsin curd test furnishes an important aid in determining any abnormal and extreme abundance of these germs and thus the blame for much of our inferior product may be located.

To recur to the lactic acid type of organisms, there can scarcely be any question that they are directly beneficial, although playing no important rôle in changing the casein into soluble products. Their general distribution and preponderance in the cheese both indicate their usefulness. Further, as we have already stated, the cheese-maker's art is an empirical attempt to produce these very germs, at least such is the case with cheddar cheese. It would be interesting to know whether they are also present in other kinds of cheese. Some analysis that we have made would seem to indicate that they are essential to many other varieties, but our data are not sufficient to warrant such conclusions. A more detailed study of the cheese flora is in progress the results of which may be expected in due time.

What then is the rôle of these acid germs? As has been suggested, they probably furnish much of the flavor, peculiar to our best cheese. The acid may give to the cheese that sharpness or "twang," so highly prized by cheese connoisseurs. Possibly, also those more delicate flavors and aromas are due to particular species of this class. However, too little is as yet known concerning this subject, to warrant any positive assertions. A careful and accurate study of the cheese flora accompanied by tests of the various germs in cheese making may lead to discoveries which when put into practice will redeem much of the inferior product now found in the market.

BULLETIN OF THE SCIENTIFIC LABORATORIES OF DENISON UNIVERSITY.
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MODIFICATION IN THE JONATHAN CREEK DRAINAGE BASIN.[1]

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My attention was first called, by a map study of the restored pre-glacial drainage of the state, to the possible existence of a preglacial drainage line extending from Zanesville, in Muskingum county, southwest along the line of Jonathan creek in the direction of the Licking reservoir. An extensive examination was made of this region during the fall of 1897, in company with Prof. W. G. Tight, of Denison University, the valley of Jonathan creek was carefully traced throughout the entire course.

The areas drained by this stream and its tributaries, as shown by the map (Plate XXV), include Newton, Harrison and Hopewell townships of Muskingum county, Clayton, Madison, Hopewell and Thorn townships of Perry county and Bowling Green township of Licking county.

The heavy broken lines represent the county limits, the small continuous lines those of the townships. Black wavy lines mark the course of the present drainage system of Jonathan creek and its tributaries. A portion of the north branch of Rush creek is shown near the bottom of the map. At the extreme upper right hand corner a section of the Muskingum river is represented, showing the entrance of the Licking river at Zanesville and that of the Moxahala river some miles below. Heavy broken lines outline the preglacial valley walls. A part of the Licking reservoir is shown to the left.

Jonathan creek rises on a broad, level, drift, plain near the eastern extremity of the Licking reservoir. At Thornport, which is situated upon the drift deposit, gas borings show a depth of drift of over 200 feet.

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[1] Accepted as a thesis in Geology in Denison University.

Rising as a small stream, winding its course over a broad alluvial deposit, Jonathan creek flows in a southeasternly direction past Glenford in Hopewell township of Perry county, through Madison township, into Newton township of Muskingum county. Eight miles below Zanesville it unites with its south branch to form the Moxahala river. The river formed at this point flows north for a distance of about 3 miles when it turns to the right through a rock gorge and empties into the Muskingum river.

The valley at Thornport is quite broad and filled up with silted material. Here the valley is 2 miles wide, and is bounded by gentle sloping hills rising to the height of 150 feet to 200 feet above the alluvial flood plain.

A view from the north side of the valley just to the east of Thornport, looking toward the west, shows the broad drift plain on which the Licking reservoir is situated extending many miles to the west. A view to the east from the same point shows the valley extending eastward and occupied by Jonathan creek. A broad valley, deeply filled with drift, its surface strewn with cat-swamps and bounded by sloping rock-walls so characteristic of the old preglacial valleys of the state.

Proceeding eastward the valley gradually narrows. The rock-walls preserve their sloping character and the creek continues to flow in a shallow trough cut through the silted deposit. At Glenford the valley has become quite narrow and 1 mile east of that place the valley is only ½ mile wide. Here is found the last of the glacial till within the valley. Just south of the town a hill with rock nucleus and capped with glacial till rises to the height of 110 feet above the flood plain of the creek. This accumulation extends off toward the northeast forming what at first sight seems to be another distinct glacial deposit, but which in reality proves to be a continuation of the same deposit, cut through and worn down by the small but erosive stream.

Numerous drift deposits occur at various levels along the valley from Thornport to Glenford, but to the east of Glenford none were observed in the immediate valley above the terrace

plain of the creek. A view from the summit of this glacial hill at Glenford, looking to the east, shows the valley still occupied by the creek, which continues to flow in a shallow course through an almost level alluvial flood plain. The valley is much narrower however than at Thornport and the walls have become quite steep.

East of Glenford a tributary from the north enters Jonathan creek. This stream has its source in Hopewell township, of Licking county, in the vicinity of Flint ridge. It is a small stream flowing in a deep, narrow valley, a form characteristic of the many tributaries.

One mile below Glenford and between the junction of this tributary and the town an outcrop of rock projects out from the north side of the valley wall. Immediately to the east of this rock promontory and on the same side of the valley, a deposit of drift material reaches an elevation of sixty feet above drainage. This deposit is composed of stratified layers of fine clays, silts, sands, gravel, flint and moranic material.

The valley of Jonathan creek filled to this level with glacial waters, washing with tremendous force against the side of the projecting rocks caused an eddy to be formed in the stream of glacial waters just below the projecting rocks. In the still waters thus produced the clays, sands and glacial gravels, carried along by the icy torrents, were deposited. The tributary from Flint ridge, flowing into this ponded water periodically deposited its load of material consisting of flinty gravels, thus aiding in building up the accumulation of heterogenious material. The interstratification of the local and glacial material is well marked. The finer quality of sand from this deposit is used by the railroad company in their locomotive sand-domes. An excavation has been made into the deposit for the laying of a track exposing a fine section.

On the north, at this point, the valley wall rises to an elevation of 200 feet. Looking eastward and southward from this elevation the view extends for many miles over an apparently broad, level cretaceous peneplain; in reality much disected by drainage channels.

Four miles to the east another tributary enters the valley from the south. Rising near Somerset in Perry county and flowing northward through Hopewell township of the same county. To the east of this southern tributary several more smaller streams flow into Jonathan creek from the north.

Near the county line between Muskingum and Perry county a very much larger tributary enters the valley from the south. This stream rises in Clayton township of Perry county, flowing northward through a part of Madison township of the same county, and enters Jonathan creek a mile and a half west of the Muskingum and Perry county line. Its valley is similar to and continuous with the valley of Jonathan creek, while just to the east the valley of Jonathan creek is very much smaller and narrower. From Thornport east Jonathan creek flows in an old valley filled up to a great depth with silt.

From a width of 2 miles at Thornport the valley gradually narrows toward the east. At Glenford the valley is about one half mile in width. The valley continues to narrow from this point on to the east and near the Muskingum and Perry county line, the valley is not over 500 feet in width. Passing east from Thornport the valley walls loose their gently sloping character and assume a more rugged and precipitous form. At Mt. Perry the valley is 300 yards wide. A short distance from this place, and almost at the county line the valley walls are 150 yards apart at flood plain. Soon the valley comes to have the appearance of a veritable gorge. The gorge extends for 3 miles with a width varying from 150 to 500 feet, bounded by rock walls, which rise to the height of 200 feet above the flood plain of the creek, as is shown on the map of the col. (Plate XXVI.)

The light continuous lines on Plate XXVI represent 50 foot contour lines measured from the flood plain. The heavy black wavy line shows the present course of Jonathan creek through the gorge; the broken line indicating its abandoned course. The Columbus, Shawnee and Hocking railroad is mapped in as a continuous line with numerous intersecting short lines. Points heavily shaded mark places where rock cuts have been made to allow for the railroad and creek beds.

In the three miles represented on the map there are seven rock cuts. Five for the railroad and two for the creek. The course of the creek through the gorge has been so changed by the railroad company that at present it runs in a much different course from that which it formerly occupied. The abandoned portions of the old channel still remain and are partially filled with water, supplied by surface waters and springs.

At the point A, at the beginning of the gorge the railroad makes a narrow cut through the rock and then parallels the creek for a short distance. At B, the rock wall projects out from the south side of the gorge and through this the railroad makes another rock cut. A new course for the creek has also been made through the rock at this point. The abandoned portion of the channel at B, has a width of 300 feet with a rock wall rising quite precipitously to an elevation of 200 feet on the north side. On the opposite side of the gorge at this point the ascent is more gradual but to an equal height. At C a cut similar to that for the railroad at B is made through the rock, projecting from the north side of the gorge. But this rock projection the creek flows in its natural course, but is soon forced to leave this and pass parallel to the railroad through a cut in the rock at D.

To the north of D the old chànnel is 250 feet in width and partially filled with water. At D, the same fact is noticeable as at B, that on the opposite side from the rock projecting into the gorge the wall rises much steeper than on the same side of the projection.

At E the creek flows in its old channel. On the south side of the gorge at this point the rock wall rises 250 feet in height at a very steep angle. The railroad again cuts through the rock at E. The creek continues to run for some distance from E in its natural course but is turned from the course before reaching F by the road bed of the railroad and flows around the rock projecting from the north side of the gorge.

The old channel between E and F has a width of 500 feet. Throughout the entire gorge Jonathan creek runs on rock bed, the walls rising to an elevation of over 200 feet on each side at

high angles. Proceeding eastward from the gorge the valley increases in width. This gorge certainly marks the position of an old col.

One half mile east of Fultonham Buckeye creek enters Jonathan creek from the south; flowing in a broad open valley with rock walls sloping gradually down to the flood plain of the contained stream. Buckeye creek rises in Harrison township, of Perry county, flows northward into Newton township of Muskingum county, where it joins Jonathan creek.

At the entrance of the valley from the south a broad gravel terrace stretches across Jonathan creek valley and extends up the valley of Buckey creek. This terrace has an elevation of 25 feet above drainage. Jonathan creek has cut its way down through this deposit and is now running on a rock floor at this point. From the junction of Buckeye creek west to the col, Jonathan creek valley is narrow and gorge-like in appearance. To the east from this point the valley is broader and gives evidence of being a direct continuation of Buckeye creek valley.

From Newtonville to the Moxahala river the valley is filled in many places with gravel terraces.

The south fork of Jonathan creek rises in the southeastern part of Perry county, receiving numerous tributaries it flows north through a broad open valley to its junction with its west branch below the village of Darlington. The Moxahala river at this point flows northward in a broad valley continuous with the valley of the south fork.

At Darlington the river leaves its preglacial valley and breaking through a gorge empties into the Muskingum river three miles south of Zanesville. The abandoned valley of the Moxahala river filled up with gravel, continues north to Zanesville and opens into the Muskingum at the mouth of the Licking river.

Three miles south of Zanesville, between the buried channel of the Moxahala river and Muskingum river and just to the north of the Moxahala gorge, Observation Knob rises to the height of 260 feet above the flood plain of the Moxahala.

A view to the east from such an elevation shows the narrow valley of the Muskingum. To the west Jonathan creek valley winds its way among the carboniferous capped hills. Across the Muskingum numerous smaller valleys open toward the river with northward direction of discharge To the northwest the valley of the Licking is outlined.

As indicated on the map, Plate XXV, broken crossed lines trace the divides through a portion of the country represented.

In the western part of Perry county a divide separates the Jonathan creek drainage in Hopewell and Thorn townships from the Rush creek drainage. Almost at right angles to this, another divide running through Reading and Clayton townships of Perry county, and continuing down into Morgan county, separates the drainage of Jonathan creek from that of the east branch of Rush creek. The C. and M. railroad tunnels through this divide near New Lexington, as does the B. and O. near Somerset.

Beginning in Hopewell township of Licking county a divide runs southeast through Hopewell township of Muskingum county. Following the county line between Muskingum and Perry counties it continues south into Clayton and Harrison townships of Perry county, intersecting the Rush creek divide near New Lexington. It is in this divide that the Jonathan creek col is located.

Let us now look for some explanation of the topography of Jonathan creek valley. The whole succession of events accompanying the first advance and final recession of the great ice sheet is considered and an explanation attempted on such a basis. Before the advance of the ice sheet, whose front extended to the north and south line through Licking and Perry counties, represented on the map, Plate XXV, with a lobe of ice running up the Jonathan creek valley as far as Glenford, Jonathan creek comprised two small streams heading on the opposite side of a north and south divide. The stream flowing to the west rising in Clayton township of Perry county, cours-

ing northwest past Glenford, emptied into the old preglacial Muskingum valley near the Licking reservoir.[1]

The other stream on the east side of the divide flowed east past the Moxahala gorge through the now buried channel of the Moxahala river and emptied into the preglacial north-flowing Muskingum at Zanesville.

When the ice sheet invaded Ohio, covering the central and western parts of the State, that portion of Jonathan creek flowing from the western side of the divide was filled with glacial waters. Its current was reversed and the great volume of water poured down from the tongue of ice extending into the valley, ponded up against the divide. At the point indicated by the col on the map, the glacial waters cut through the divide, uniting the two drainage systems into one present, continus system. The geological structure of the divide amply allows for the consideration of such a theory. The narrowness of the valley immediately to the east of the col, the silting up of the valley from the col west to Glenford, the furrowed out character of the valley east of the col all lead to such a conclusion.

Attention should be called to the form of the section of the valley east of the gorge. At the top it has the appearance of the old preglacial valleys: broad, with sloping sides. The lower portion, however, is deeply furrowed out, evidently the result of recent water action. Before the torrents of glacial waters poured over the divide, the stream then occupying this portion of the valley, coursed through a broad open valley. The volume of water which came down from the ice field, loaded with eroded material, cut down the old valley to a great depth, leaving it in its present form.

The preglacial channel of the Moxahala river from where it leaps through the divide, between the south fork of Jonathan creek and the Muskingum river to Zanesville, has a depth of filling of 50 feet above the flood plain of the Moxahala.

When the rivers of glacial waters forced the north-flowing preglacial Muskingum to change its direction of flow, the coun-

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[1] Bulletin Vol. VIII, Pt. 2.

ter currents sent up the old valley checked the flow of the Moxahala river then coursing through it. In the backed up waters the great accumulations of gravel now filling the valley were deposited. Too strong a current was sent up the old valley for the Moxahala river to stem. As a result the Moxahala broke over the divide existing between the south fork and the Muskingum river, and finally cut its course down to the present level.

After the recession of the ice sheet these cols remained in use, having been worn down to a sufficient depth.

These conclusions in reference to Jonathan creek are in harmony with the restored preglacial drainage of the neighboring territory and thus aid in establishing the truthfulness of those restorations.

The south fork of Jonathan creek in its northern direction to Zanesville conforms to the north flowing Muskingum. The west fork of Jonathan creek, from the col, westward to the Licking reservoir, formed a lateral tributary to the preglacial Muskingum in its course from Dresden Junction through the old abandoned valley to Newark and thence southwestward past the Licking reservoir on its way to the Scioto basin.

With these references it is hoped that one more item, though small it may be, is added to the knowledge of the restored preglacial drainage of Ohio.

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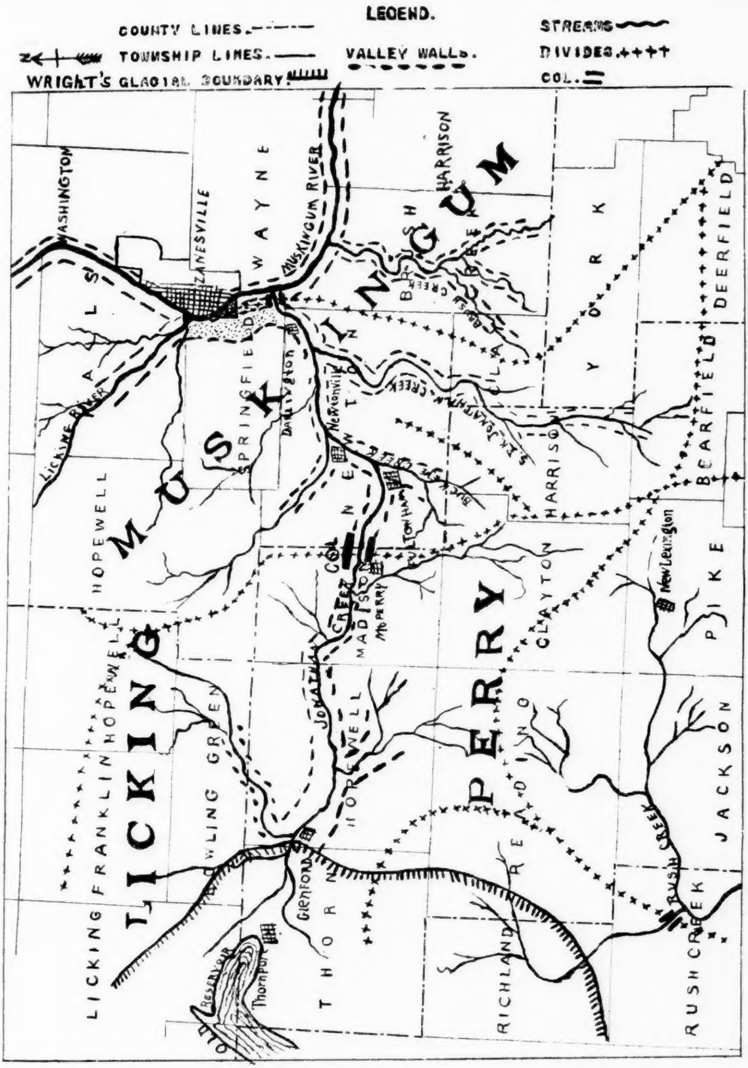
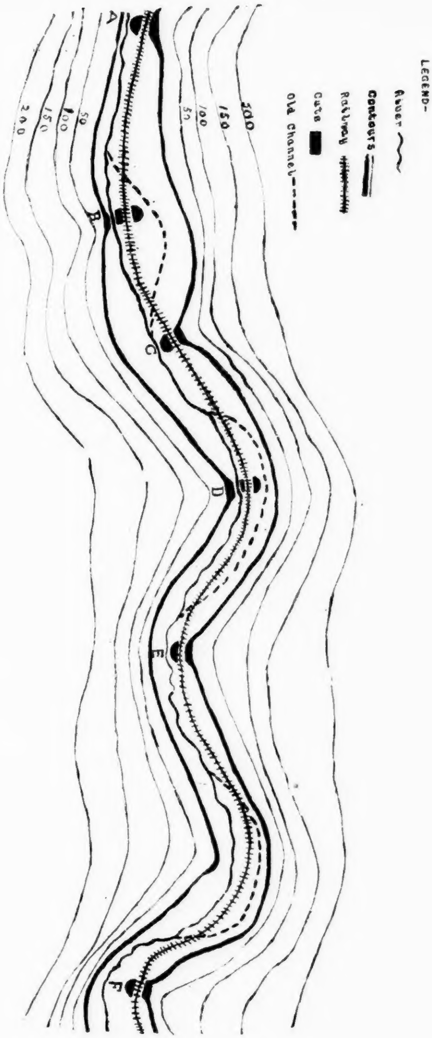


JONATHAN CREEK DRAINAGE BASIN

DAVIS—Jonathan Creek Drainage Basin.



JONATHAN CREEK COL.

DAVIS—Jonathan Creek Drainage Basin.